#14/30
1/27/01

PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant: Maurice Cohen et al..

Serial No.: 09/250,883

Filed: February 16, 1999

For: REAGENTS AND METHODS USEFUL FOR DETECTING DISEASES OF THE BREAST

Examiner: C. Myers

Group Art Unit: 1655

Case No.: 6131.US.C1

Date: January 4, 2001

CERTIFICATE OF MAILING (37 CFR 1.8 (a))

I hereby certify that this paper (along with any paper referred to as being attached or enclosed) is being deposited with the United States Postal Service on the date shown below with sufficient postage as first class mail in an envelope addressed to the:

Assistant Commissioner for Patents
Washington, D.C. 20231, on:

Date of Deposit: January 11, 2001

Wanda E. Smith 1/11/01
Wanda E. Smith Date

DECLARATION OF PAULA N. FRIEDMAN Ph.D.

Assistant Commissioner for Patents
Washington, D.C. 20231

Dear Sir:

1. I am one skilled in the art of cancer diagnostics. I have a Ph.D. in Molecular Biology from Columbia University as well as an M.A. and a M. Phil. in Molecular Biology also from Columbia University. I further have a B.A. in Biology from Dartmouth College.

2. I was a Postdoctoral Fellow in the Laboratory of Dr. Clay Siegall at the Pharmaceutical Research Institute Bristol–Myers Squibb and an Assistant Pharmacologist, Dept. of Clinical Immunology & Biol. Therapy at the MD Anderson Cancer Center.

3. I have nine years of research and development experience in the cancer diagnostic industry. Much of my work has involved the discovery and validation of novel cancer markers to improve the accuracy of diagnosing the onset of cancer. In fact, I am a named inventor of several U.S. Patents, all of which are related to the field of cancer diagnostics.

4. I also have authored numerous journal articles relating to cancer pathology, detection, and metastasis (see Attachment I).

5. I am one of the named inventors of the aforementioned application.

6. I have read and am familiar with the Patent Office Action dated September 13, 2000 and utility rejection under 35 U.S.C. 101 applied against the present application.

7. I have reviewed the data illustrating CEA and PSA tissue specificity generated using the Incyte Lifeseq Gold database (Attachment II), the same database utilized to generate Example 1 in the instant application.

8. CEA is a tissue-specific marker that has been shown to be highly expressed in the GI tract. As evidenced by Attachment II, 61 out of 148 GI libraries express CEA whereas only 27 out of 1,144 other, non-GI tract libraries express this gene. Therefore, CEA is expressed approximately 17 times more in GI tissue when compared to the rest of the body.

9. Similarly, PSA is a tissue-specific marker that has been shown to be highly expressed in the prostate. As evidenced by Attachment II, 65 out of 79 prostate libraries (classified as male genitalia) express PSA whereas only 22 out of 1213 other, non-prostate libraries express this gene. Therefore, PSA is expressed approximately 45 times more in prostate when compared to the rest of the body. Further, the PSA gene product is utilized in screening, prognosis, and monitoring prostate cancer patients by oncologists and it is recommended that all men over the age of 40 be tested yearly with a PSA assay.

10. To those skilled in the art, such as myself, PSA and CEA are well known tumor markers, which indicate cancer of the prostate (PSA) and gastrointestinal (GI) tract (CEA) when the respective gene product is found in the blood sample of a patient.

11. As shown in the instant specification, (Example 1, p.52, starting on line 20) BS203 is 22 times more abundant in breast tissue than non-breast tissue.

12. Clearly, BS203 is characteristic of a tissue specific marker and able to act as a cancer diagnostic, as evidenced by the above data.

13. Tissue-specific markers such as PSA and CEA are the most diagnostic tools in early oncology detection and are used on a daily basis.

14. Based on the statistics in the Incyte database, BS203 is clearly a breast specific marker and, therefore, its use as a breast cancer marker is unquestionable.

15. I hereby declare that all statements made herein are of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that the statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under § 1001 of Title 18 of the United States code and such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Paula N. Friedman, Ph.D.

1/5/01
Date

ATTACHMENT I

Publications:

Wang, E.H., **Friedman, P.N.** and Prives, C. 1989. The murine p53 protein blocks replication of SV40 DNA in vitro by inhibiting the initiation functions of SV40 large T antigen. Cell, 57, 379-392.

Manfredi, J.J., Wang, E.H., **Friedman, P.N.,** and Prives, C. 1989. Purified SV40 large T antigen in complex with murine p53 does not support SV40 DNA replication in vitro. Implications for the mechanism of transformation by SV40 large T antigen. In Common Mechanisms of Transformation by Small DNA Tumor Viruses. Luis P. Villeareal, ed. American Society for Microbiology,113.

Bischoff, J.R., **Friedman, P.N.,** Marshak, D., Prives, C., and Beach, D. 1990. The p53 protein is phosphorylated by cyclin A-cdc2 as well as cyclin B-cdc2. PNAS, 87,4766-4770.

Friedman, P.N., Kern, S.E., Vogelstein, B., and Prives, C. 1990. Wild-type, but not mutant, human p53 proteins inhibit the replication activities of simian virus 40 large tumor antigen. PNAS, 87, 9275-9279.

Kern, S.E., Kinzler, K.W., Baker, S.J., Nigro, J.M., Rotter,V., Levine, A.J., **Friedman, P.N.,** Prives, C. and Vogelstein, B. 1991. Mutant p53 proteins bind DNA abnormally in vitro. Oncogene, 6, 131-136.

Bargonetti, J., **Friedman, P.N.,** Kern, S.E., Vogelstein, B., and Prives, C. 1991. Wild-type but not mutant p53 immunopurified proteins bind to sequences adjacent to the SV40 origin of replication. Cell, 65, 1083-1091.

Kern, S.E., Kinzler, K.W., Bruskin, A., Jarosz, D., **Friedman, P.N.,** Prives, C. and Vogelstein, B. 1991. Identification of p53 as a sequence-specific DNA-binding protein. Science, 252,1708-1710.

Farmer, G., Bargonetti, J., Zhu, H., **Friedman, P.N.,** Prywes, R., and Prives, C. 1992. Wild-type p53 activates transcription in vitro. Nature, 358, 83-86.

Prives, C., Bargonetti, J., **Friedman, P.N.,** Manfredi, J.J., and Wang, E. 1992. Functional consequences of the interaction between the p53 tumor suppressor protein and the SV40 large tumor antigen. Cold Spring Harbor Symposium on Quantitative Biology: The Cell Cycle, Vol. 56, 227-235.

Bargonetti, J., Reynesdottir, I., **Friedman, P.N.,** and Prives, C. 1992. Wild-type p53 site-specific binding to cellular DNA is regulated by SV40 T antigen and mutant p53. Genes and Devel., 6, 1886-1898.

Friedman, P.N., Wang, E.H., Meerovitch, K., Sonenberg, N., and Prives, C. 1992. Murine p53 inhibits the function but not the formation of SV40 T antigen hexamers and stimulates T antigen RNA helicase activity. Chromosoma, 102, 60-66.

Friedman, P.N., Chen, X., Bargonetti, J., and Prives, C. The p53 protein is an unusually shaped tetramer that binds directly to DNA. PNAS, 90, 3319-3323.

Reynesdottir, I., Lorimer, H.E., **Friedman, P.N.,** Wang, E.H., and Prives, C. 1993. Phosphorylation and active ATP hydrolysis are not required for SV40 T antigen hexamer formation. J. of Biol. Chem., 268, 24647-24654.

Friedman, P.N., McAndrew, S.J., Gawlak, S.L., Chace, D., Trail, P.A., Brown, J.P., and Siegall, C.B. 1993. BR96 sFv-PE40, a potent single-chain immunotoxin that selectively kills carcinoma cells. Cancer Res., 53, 334-339.

Friedman, P.N., Chace, D.F., Trail, P.A., and Siegall, C.B.1993. Antitumor activity of the single-chain immunotoxin BR96 sFv-PE40 against established breast and lung tumor xenografts. J. of Immun., 150, 3054-3061.

Friedman, P.N., Chace, D.F., Gawlak, S.L., and Siegall, C.B. 1993. The single-chain immunotoxins BR96 sFv-PE40 and BR96 sFv-PE38: Potent anti-tumor agents for the treatment of human cancer. In Growth Factors, Peptides, and Receptors, T. Moody, ed., Plenum Press, 409-414.

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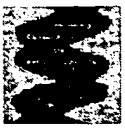
ATTACHMENT II
Gene Detail



Main Menu | Genes | Libraries | Transcripts | Library Comp | Genomic Data | Sequences | Help

Gene Information

Gene ID: 396578 Template Count: 6 Seq Count: 557 Clone Count: 483
Top Hit ID: g180198 Hit Species: Homo sapiens
Hit Type: Exact E-Value: 0.0e+00 Pct ID: 99

Gene Description: Human carcinoembryonic antigen mRNA.

Genomic Data

Templates

		Template ID	Template Description	Top Hit ID	Hit Species
☑		396578.12	Human carcinoembryonic antigen mRNA.	g180198	Homo sapien
☑		396578.6	Human BGP gene for biliary glycoprotein,	g29447	Homo sapien
☑		396578.10	Human carcinoembryonic antigen (CEA) gen	g180206	Homo sapien
☑		396578.15	Human carcinoembryonic antigen mRNA (CEA	g180222	Homo sapien
☑	●	396578.2	Human carcinoembryonic antigen mRNA.	g180198	Homo sapien
☑		396578.14	Human carcinoembryonic antigen (CEA) gen	g180208	Homo sapien

Uncheck All

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Protein Function

Function hierarchy
- Adhesion and molecular recognition
  - Adhesion
    - Ig superfamily
- Localized and structural proteins
  - Membrane
    - Peripheral and anchored membrane proteins

Tissue Distribution

Tissue Category	Clone Count	Found in	Abs Abund	Pct Abund	Pct Spec
Cardiovascular System	266190	0/68	0	0.0000	0.00
Connective Tissue	144645	1/47	1	0.0007	0.64
Digestive System	501101	61/148	342	0.0682	62.17
Embryonic Structures	106713	0/21	0	0.0000	0.00
Endocrine System	225386	1/53	1	0.0004	0.36
Exocrine Glands	254635	2/64	3	0.0012	1.09
Genitalia, Female	427284	8/106	17	0.0040	3.65
Genitalia, Male	448207	2/114	5	0.0011	1.00
Germ Cells	38282	0/5	0	0.0000	0.00
Hemic and Immune System	680277	4/159	49	0.0072	6.56
Liver	109378	1/35	2	0.0018	1.64
Musculoskeletal System	159280	0/47	0	0.0000	0.00
Nervous System	955753	0/198	0	0.0000	0.00
Pancreas	110207	4/24	26	0.0236	21.51
Respiratory System	390086	4/93	6	0.0015	1.37
Sense Organs	19256	0/8	0	0.0000	0.00
Skin	72292	0/15	0	0.0000	0.00
Stomatognathic System	12923	0/10	0	0.0000	0.00
Unclassified/Mixed	120926	0/13	0	0.0000	0.00
Urinary Tract	279062	0/64	0	0.0000	0.00
Totals	5321883	88/1292	452	0.0091	100.00

Requested by: LG051_092000_GOLD_RO on Thu Sep 28 14:59:30 2000

IncyteGenomics

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Gene Detail

Gene Information

Gene ID: 1383133 Template Count: 17 Seq Count: 2876 Clone Count: 279
Top Hit ID: g35740 Hit Species: Homo sapiens
Hit Type: Exact E-Value: 0.0e+00 Pct ID: 99

Gene Description: Human mRNA for prostate specific antigen.

Genomic Data

Templates

		Template ID	Template Description	Top Hit ID	Hit Species
☑		1383133.7	Human mRNA for prostate specific antigen	g35740	Homo sapien
☑	•	1383133.17	Human mRNA for prostate specific antigen	g35740	Homo sapien
☑		1383133.8	Human mRNA for prostate specific antigen	g35740	Homo sapien
☑		1383133.16	Human prostate specific antigen (PA) gen	g189525	Homo sapien
☑		1383133.13	Human mRNA for prostate specific antigen	g35740	Homo sapien
☑		1383133.2	Human PSA gene for prostate specific ant	g35730	Homo sapien
☑		1383133.9	Human prostate-specific antigen (PA) gen	g341200	Homo sapien
☑		1383133.11	Human mRNA for prostate specific antigen	g35720	Homo sapien
☑		1383133.14	Human mRNA for prostate specific antigen	g35740	Homo sapien
☑		1383133.12	Human prostate specific antigen gene, co	g190552	Homo sapien
☑		1383133.15	Human prostate specific antigen gene, co	g190552	Homo sapien
☑		1383133.10	Human prostate specific antigen gene, co	g190552	Homo sapien
☑		1383133.1	Human prostate specific antigen (PA) gen	g189523	Homo sapien
☑		1383133.3	Human prostate specific antigen (PA) gen	g189529	Homo sapien
☑		1383133.4	Human mRNA for prostate specific antigen	g35740	Homo sapien
☑		1383133.5	Human mRNA for prostate specific antigen	g35720	Homo sapien
☑		1383133.6	Human mRNA for prostate specific antigen	g35740	Homo sapien

Uncheck All

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Protein Function

Enzyme hierarchy
- Hydrolases
  - Acting on peptide bonds (peptide hydrolases)
    - Serine endopeptidases

Function hierarchy
- Protein modification and maintenance
  - Proteases
    - Serine proteases
- Localized and structural proteins
  - Secreted and extracellular
    - Other secreted and extracellular

Pathway hierarchy
- Metabolism
  - Protein metabolism
    - Post-translation
      - Protein cleavage and degradation

Tissue Distribution

Tissue Category	Clone Count	Found in	Abs Abund	Pct Abund	Pct Spec
Cardiovascular System	266190	0/68	0	0.0000	0.00
Connective Tissue	144645	1/47	4	0.0028	0.45
Digestive System	501101	6/148	6	0.0012	0.19
Embryonic Structures	106713	1/21	1	0.0009	0.14
Endocrine System	225386	1/53	1	0.0004	0.06
Exocrine Glands	254635	3/64	4	0.0016	0.26
Genitalia, Female	427284	1/106	1	0.0002	0.03
Genitalia, Male	448207	65/114	2728	0.6086	97.07
Germ Cells	38282	0/5	0	0.0000	0.00
Hemic and Immune System	680277	1/159	5	0.0007	0.11
Liver	109378	0/35	0	0.0000	0.00
Musculoskeletal System	159280	2/47	13	0.0082	1.31
Nervous System	955753	5/198	6	0.0006	0.10
Pancreas	110207	1/24	2	0.0018	0.29
Respiratory System	390086	0/93	0	0.0000	0.00
Sense Organs	19256	0/8	0	0.0000	0.00
Skin	72292	0/15	0	0.0000	0.00
Stomatognathic System	12923	0/10	0	0.0000	0.00
Unclassified/Mixed	120926	0/13	0	0.0000	0.00
Urinary Tract	279062	0/64	0	0.0000	0.00
Totals	5321883	87/1292	2771	0.0524	100.00

Requested by: LG051_092000_GOLD_RO on Thu Sep 28 15:06:43 2000